THE UNIVERSITY OF KANSAS
SCIENCE BULLETIN
VOL. XLI] DECEMBER 23, 1960 [NO. 8

Lung-Flukes of Snakes, Genera *Thamnophis* and *Coluber*, in Kansas

BY
PEGGY LOU STEWART

ABSTRACT: *Thamnophis sirtalis parietalis* Say and *Coluber constrictor flaviventris* Say were trapped and examined for lung-flukes on the University of Kansas Natural History Reservation, in Douglas County, in the months of June through November, 1958. These snakes were released at the points of capture after the flukes were removed. Also, examinations were made of preserved specimens of *T. sirtalis parietalis* Say, *T. radix haydeni* Kennicott, *T. sauritus proximus* Say, and *C. constrictor flaviventris* in the Museum of Natural History, the University of Kansas.

Crow (1913) reported *Natrix rhombifera, Ancistrodon* (*Agkistrodon*) *contortrix, A. mokassen* (= *A. c. mokeson*), *A. piscivorus,* and *Sistrurus miliarus* as hosts of lung-flukes in Kansas.

REVIEW OF THE TAXONOMY OF SOME TREMATODES IN REPTILES

Members of the order Digenea von Beneden are characterized by having the mouth opening within the oral sucker and an endoparasitic life in vertebrates. The members of the suborder Prosostomata Odhner, characterized primarily by having a subterminal mouth, are the only representatives of the Digenea that are parasitic in reptiles.

Yamaguti (1958) reviewed 29 families in Prosostomata, of which one, the Plagiorchiidae (Luhe, 1900) Ward (1917), comprises flukes having the distinctive character of tandem, diagonal, or symmetrical testes in the posterior half of the body.

Pratt (1902) defined the subfamily Reniferinae, in the family Plagiorchiidae. Pratt assigned to the Reniferinae those genera having the genital pore lateral to a point midway between the suckers: *Styphlodora* Looss (1899), *Astiotrema* Looss (1900), *Renifer* Pratt (1902), *Ochetosoma* Braun (1901), and Oistosomum Odhner (1902). Baer (1924) raised the Reniferinae to family level, Reniferidae.

Talbot (1934) included the following seven genera in the Reniferinae: *Macrodera* Looss (1899), *Renifer, Lechriorchis* Stafford (1905), *Zeugorchis* Stafford (1905) *Pneumatophilus* Odhner (1910), *Dasymetra* Nicoll (1911), and *Caudorchis* Talbot (1933).

Mehra (1937) placed in the subfamily Reniferinae sixteen genera, including *Macrodera, Renifer, Lechriorchis, Zeugorchis, Pneumatophilus, Natriodera* Mehra (1937), and *Pseudorenifer* Price (1936), especially pertinent to my discussion. He made use of the miracidia, sporocysts, and cercariae in assigning the mentioned genera to one subfamily. Adults of species in the Reniferinae, as recognized by Mehra, develop from xiphidiocercariae.

Byrd and Denton (1938) rediagnosed the subfamily Reniferinae and considered the following eight genera as members of the subfamily: *Renifer, Lechriorchis, Zeugorchis, Pneumatophilus, Dasymetra, Natriodera, Neorenifer* Byrd and Denton (1938), and *Paralechriorchis* Byrd and Denton (1938).

The genera *Styphlodora, Astiotrema, Ochetosoma,* and *Oistosomum* were excluded from the Reniferinae by Byrd and Denton, who agreed with Talbot (1934) that the positions of the genital pore and cirrus pouch of these genera exclude them from the subfamily as it was defined by Baer (1924).

Byrd and Denton (1938) opined that the subfamily Reniferinae should include genera having the genital pore in a position lateral to the digestive ceca or the pharynx. One genus, *Renifer,* including *R. ellipticus* Pratt (1902), is limited by these authors to those species having the genital pore lateral to the bifurcated ceca. *Renifer elongatus* Pratt (1903), *Renifer kansensis* Crow (1913), and *Renifer aniarum* Leidy (1891) are excluded because the genital pore is lateral to the oral sucker and pharynx.

Price (1936) concluded, after examining and redescribing Stafford's specimens in the genus *Zeugorchis* Stafford (1905) and in *Lechriorchis* Stafford (1905), that *Caudorchis* Talbot (1933) is synonymous with *Zeugorchis.* Price further considered *Zeugorchis bosci* Cobbold (1859), *Z. syntomentera* Sumwalt (1926), and *Z. megametricus* Talbot (1934) not to be congeneric with the type species *Z. aequatus* Stafford (1905). *Z. bosci* has the genital pore in a comparable position and ceca of approximately the same length as those members of the genus *Dasymetra* Nicoll (1911). Similar variables, such as genital pore posterior to or to one side of the bifurcation of ceca, are found in the other three species. Price (1935) established a new genus, *Pseudorenifer,* for *megametricus, ancistrodontis,* and *syntomentera,* whose anatomical features do not agree

with those of *Z. aequatus*. *Pseudorenifer megametricus* Talbot (1934) was designated as the type species of the new genus.

According to Byrd and Denton (1938), *Zeugorchis* is characterized by the position of the genital pore, which is immediately posterior to the bifurcation of the ceca; and by the position of the testes, which are situated in the posterior fourth of the body. Since Price designated *megametricus* as the type species of the genus *Pseudorenifer*, in which the genital pore is lateral to the bifurcation of the ceca, Byrd and Denton consider the genera *Pseudorenifer* and *Renifer* to be synonymous.

For two groups of species that could not be placed in any named genus, Byrd and Denton (1938) proposed two new genera: *Paralechriorchis* comprising those species having the genital pore confined to the area between the acetabulum and the bifurcation of the ceca, and *Neorenifer* comprising those species having the genital pore at or near the margin of the body and in the region of the oral sucker and pharynx. The species *Renifer elongatus* Pratt, *Renifer kansensis* Crow and *Renifer aniarum* Leidy, among others, were assigned to the new genus *Neorenifer*.

DESCRIPTION OF NEW SPECIES

Family RENIFERIDAE Baer, 1924

Subfamily Renieferinae Pratt, 1902

Genus *Zeugorchis* Stafford, 1905

Zeugorchis megacystis, new species

(Pl. 1, Pl. 2)

Holotype.—No. 39104 U. S. National Museum, host *Thamnophis sirtalis parietalis* Say 5¾ mi. N. E. Lawrence, Douglas Co., Kansas; obtained on June 10, 1958, by Peggy Lou Stewart.

Paratypes.—No. 39106 (four individuals), U. S. National Museum, host species and locality of capture same as for holotype; obtained on June 10, July 7, 19, and 30, all in 1958.

Diagnosis.—Body attenuated anteriorly, length 2.5 mm., width 0.66 mm. (relaxed living example); entire cuticula randomly spinose; oral sucker subterminal; acetabulum immediately anterior to middle of body, larger than oral sucker; prepharynx shorter than pharynx; esophagus longer than combined length of prepharynx and pharynx; intestinal ceca extending slightly beyond posterior extremity of acetabulum; genital pore median to right cecum, posterior to anterior extremity of acetabulum; cirrus pouch narrowly

ovoid, larger posteriorly; testes oval, in posterior third of body; ovary oval, overlapping posterior margin of acetabulum; uterus convoluted; excretory bladder ventral to uterus; vitellaria lateral, extending more anteriorly than posteriorly from middle of body; ova numerous, yellow-brown, 0.02 to 0.05 mm. by 0.01 to 0.02 mm; parasitic in digestive and respiratory tracts of *Thamnophis sirtalis parietalis, T. radix haydeni,* and *T. sauritus proximus.*

Measurements of holotype and variations in corresponding measurements of paratypes.*

	Length	Width	Thickness
Body	2.63±0.35(2.54)	0.72±0.06(0.66)	0.22±0.02
Oral sucker	0.35±0.01(0.45)	0.30±0.02(0.35)	0.21±0.02
Acetabulum	0.41±0.07(0.46)	0.46±0.01(0.48)	0.09±0.01
Prepharynx and pharynx	0.06±0.01(0.14)	0.15±0.01(0.14)	0.11±0.02
Esophagus	0.16±0.02(0.22)	0.12±0.01(0.06)	0.10±0.01
Ceca	0.79±0.03(0.72)	0.06±0.02(0.05)	0.02±0.01
Genital pore	.....	0.01±0.01(0.01)	0.02±0.01
Cirrus pouch	0.34±0.01(0.40)	0.12±0.01(0.16)	0.06±0.01
Testes	0.02±0.02(0.21)	0.09±0.03(0.05)	0.16±0.03
Sperm duct	1.34±0.05(....)	0.04±0.01(....)	0.03±0.01
Ovary	0.09±0.01(0.09)	0.05±0.02(0.05)	0.10±0.02
Uterus	.....	0.25±0.16(0.42)	0.08±0.04
Excretory bladder	0.27±0.01(0.28)	.....	.....
Excretory tube	.....	0.02±0.02(....)	0.02±0.02
Vitelline span	0.87±0.03(0.93)	.....	.....

* Measurements of holotype are in parentheses. Dimensions are in millimeters.

Comparisons.—Among named kinds of *Zeugorchis, Z. megacystis* most closely resembles Z. *eurinus* Talbot (1933). From Z. *eurinus, Z. megacystis* differs as follows: length of body averaging approximately 2.5 *versus* 2.4 mm.; vitellaria in larger part anterior instead of posterior to middle of body; oral sucker smaller than, instead of approximately same size as, acetabulum. From *Zeugorchis syntomenteroides* Parker (1941), *Z. megacystis* differs as follows: body attenuate anteriorly instead of broad; length of body averaging approximately 2.5 *versus* 2.0 mm.; genital pore on same vertical plane as anterior margin of acetabulum instead of farther anteriorly immediately posterior to bifurcation of ceca; testes in posterior third rather than middle third of body; ovary oval instead of spherical. From *Zeugorchis aequatus* Stafford (1905), the type species of the genus, *Z. megacystis* differs as follows: body attenuate anteriorly instead of broad; length of body averaging approximately 2.5 *versus* 1.0 mm.; acetabulum larger than oral sucker instead of

smaller than oral sucker; ceca extending to posterior edge of acetabulum instead of extending immediately anterior to testes; genital pore posterior to anterior extremity of acetabulum instead of directly posterior to bifurcation of ceca; testes in posterior third instead of posterior fifth of body; ovary overlapping posterior margin of, instead of posterior to, acetabulum; vitellaria extending more anteriorly than posteriorly instead of equally anteriorly and posteriorly from middle of body.

Genus *Neorenifer* Byrd and Denton, 1938

Neorenifer lateriporus, new species

(Pl. 1, Pl. 2)

Holotype.—No. 39105 U. S. National Museum, host *Coluber constrictor flaviventris* Say; 5¾ mi. N. E. Lawrence, Douglas Co., Kansas; obtained on September 8, 1958, by Peggy Lou Stewart.

Paratypes.—No. 39107 (three individuals) U. S. National Museum, host species and locality of capture same as for holotype; obtained on June 9, July 25, and August 12, all in 1958.

Diagnosis.—Body slightly attenuated anteriorly and posteriorly; length 5.24 mm., width 0.83 mm. (relaxed living example); cuticula without spines; oral sucker subterminal; acetabulum immediately anterior to middle of body, larger than oral sucker; prepharynx absent; pharynx longer than esophagus, intestinal ceca extending slightly posterior to anterior extremity of testes; genital pore lateral

Measurements of holotype and variations in corresponding measurements of paratypes.**

	Length	Width	Thickness
Body	5.84±0.16(5.24)	1.43±0.27(0.83)	0.30±0.02
Oral sucker	0.36±0.04(0.27)	0.44±0.18(0.31)	0.22±0.17
Acetabulum	0.42±0.02(0.41)	0.48±0.05(0.43)	0.10±0.04
Pharynx	0.25±0.03(0.23)	0.41±0.12(0.25)	0.34±0.10
Esophagus	0.27±0.09(0.22)	0.14±0.12(0.06)	0.14±0.10
Ceca	2.04±0.16(1.92)	0.08±0.04(0.08)	0.04±0.03
Genital pore	0.02±0.01(....)	0.14±0.01(0.13)	0.14±0.01
Cirrus pouch	1.23±0.01(0.88)	0.16±0.01(0.14)	0.08±0.01
Testes	0.53±0.07(0.46)	0.32±0.11(0.25)	0.20±0.09
Ovary	0.39±0.04(0.16)	0.24±0.17(0.15)	0.17±0.08
Uterus	0.71±0.65(....)	0.21±0.06(0.59)	.....
Excretory bladder	1.48±0.05(1.32)	.....	.....
Excretory tube	.....	0.08±0.02(....)	0.08±0.02
Vitelline span	1.51±0.06(1.32)	.....	.....

** Measurements of holotype are in parentheses. Dimensions are in millimeters.

on right side of body; cirrus pouch narrowly ovoid, larger posteriorly than elsewhere, ending anterior to acetabulum; ovary lobed; testes oval, immediately posterior to middle of body; uterus convoluted; excretory bladder ventral to uterus; vitellaria lateral, extending more anteriorly than posteriorly from middle of body; shell gland not distinct; ova numerous, brown, 0.02 to 0.05 by 0.02 mm.; parasitic in digestive and respiratory tracts of *Coluber constrictor flaviventris.*

Comparisons.—Of the other 13 species in the genus *Neorenifer, N. orula* Talbot (1934), the type species of the genus, seems to be the closest relative of *N. lateriporus.* From *N. orula, N. lateriporus* differs as follows: genital pore on right instead of left margin of body; body less attenuated posteriorly; length averaging 5.8 *versus* 2.0 mm.; ceca overlapping anterior margin of testes instead of terminating posterior to testes; testes in middle, not posterior, third of body; ovary lobed instead of spherical; host, *Coluber constrictor flaviventris* instead of *Natrix sipedon.* The absence, instead of presence, of cuticular spines in *N. lateriporus* is the principal difference between it and *N. sauromates* Poirier (1885), *N. zschokkei* Volz (1899), *N. elongatus* Pratt (1903), *N. formosum* Nicoll (1911), *N. validus* Nicoll (1911), *N. kansensis* Crow (1913), *N. wardi* Byrd (1936), and *N. heterodontis* Byrd and Denton (1938). A difference from *N. acetabularis* Crow (1913) and *N. aniarum* Leidy (1891) is the absence instead of presence of a dichotomous pattern of the vitelline follicles in an anteroposterior plane. Differences from *N. serpentis* Schmidt and Hubbard (1940) are: body fusiform instead of ovoid; length averaging 5.8 instead of 2.8 mm.; shell gland indistinct instead of distinct; uterus convoluted instead of non-convoluted; host, *Coluber constrictor flaviventris* instead of *Agkistrodon piscivorus.* Differences from *N. septicus* MacCallum (1921) are: longer (5.8 *versus* 3.3 mm.); ceca extending beyond level of ovary instead of to level of ovary; uterus convoluted instead of non-convoluted; excretory pore opening at posterior extremity, rather than anterior to posterior end, of body.

INFECTION AND NATURAL HISTORY OF SNAKES EXAMINED

Thamnophis.—Ninety-eight living *T. sirtalis parietalis* on the reservation (June through November, 1958) and 326 preserved *Thamnophis sirtalis parietalis, T. radix haydeni, T. sauritus proximus* in the museum collection (1899 through 1957) were examined for fluke infections. Thirty-three *Zeugorchis megacystis* were found in a total of 17 of the snakes on the reservation, and 26 in a total of 6

of the snakes in the museum. Five was the largest number of flukes found in a single live snake, and 8 was the largest number found in a preserved snake.

Adult individuals of *Zeugorchis megacystis* were taken from the digestive tracts of the snakes on the reservation, and from the digestive and respiratory tracts of the snakes in the museum.

The majority of lung-flukes that were obtained from living and preserved snakes came from specimens collected in June and July.

Of the living snakes, 4 of 39 males and 13 of 59 females were infected by *Zeugorchis megacystis*.

Lengths and weights of infected and noninfected snakes were measured on the reservation. Because of the small number of infected snakes, the size-differences between individual snakes could not certainly be attributed to the effects of parasitism.

More than one hundred double traps were distributed in various habitats (including forest and prairie) on the reservation, in May. In the autumn, all of the traps were moved to rock ledges. None of the snakes trapped on the ledges was infected.

In the museum collection, trematodes were found in two snakes from Douglas County and in one each from Anderson, Stafford, Wallace and Morton counties. The specimens studied represented 55 counties, with the majority having been collected in eastern, south-central, and northwestern Kansas.

Coluber.—One hundred and twenty-three *C. constrictor flaviventris* were examined on the reservation and 294 in the museum collection. Seven of the live snakes contained a total of 13 *Neorenifer lateriporus*, whereas 19 preserved snakes were infected by a total of 122 lung-flukes. The largest number of flukes found in a single live snake was 3, in a preserved snake, 41.

The regions of infection of *N. lateriporus* in living and preserved *C. constrictor flaviventris* were the same as those of *Z. megacystis* in *Thamnophis*.

Major infections occurred in June and July in living snakes, and in preserved snakes collected in June, July, and September.

Fifty-two male and 71 female snakes were examined on the reservation; 3 males and 4 females contained lung-flukes.

Variations in length and weight of *C. constrictor flaviventris* could not be attributed to parasitism by *N. lateriporus*.

Infected and noninfected *C. constrictor flaviventris* were trapped on most of the reservation area.

Among the 26 snakes captured in Douglas County, 4 contained

N. lateriporus. Eight additional counties yielded infected snakes: Gove, seven; Clark, two; and Anderson, Graham, Neosho, Stafford, Wyandotte, one each. The study included specimens from 44 counties, with collections having been made primarily in eastern, south-central, and northwestern Kansas.

ATTEMPTS TO DISCOVER THE LIFE CYCLES OF THE FLUKES

Samples of snake scats (fecal material) were examined for ova. Eleven scat samples from *Thamnophis* contained trematode ova. These ova were of the same dimensions as ova within the uteri of *Z. megacystis.* None of the scat samples obtained from *C. constrictor flaviventris* contained trematode ova.

Snails, *Helisoma trivolvis lentum* and *Physa hawni,* were collected once a week from June 19 through July 24, 1958. Daily, over a period of six weeks, snails of the two species were macerated and examined for sporocysts and rediae. None was found.

Water samples from culture dishes containing snails were examined for cercariae, twice daily, from June 19 through July 31, 1958. The samples were negative.

The frog, *Rana pipiens* Schreber, was the primary food source for the *T. sirtalis parietalis* and *C. constrictor flaviventris* trapped on the reservation. Approximately 50 *R. pipiens* were collected near the reservation from June 26 through July 31, 1958. The frogs were dissected and examined, but no metacercariae were found.

Possible intermediate hosts found in the digestive tracts of *Coluber* and *Thamnophis* were: earthworm, *Lumbricus* sp.; voles *(Microtus);* toads, *Scaphiopus bombifrons* Cope, *Bufo terrestris* Bonaterre, and *Bufo woodhousei* Girard; frogs, *Acris gryllus* Le Conte, *Hyla versicolor* Le Conte, *Pseudacris nigrita* Le Conte, *Gastrophryne olivacea* Hallowell, and young *Rana catesbeiana* Shaw.

During dry weather, the frogs and toads inhabited the edges of the two reservation ponds; in wet weather they approached uniform dispersal throughout the reservation. Rainfall in the months of 1958 when collections were made was great enough to afford ample opportunity for the snakes to capture intermediate hosts of lung-flukes at or near all of the trap sites (assuming toads or frogs act as hosts for *Z. megacystis* and *N. lateriporus*).

In snakes examined from the reservation, no parasites (of body-part checked) were found, other than the two kinds described herein.

SUMMARY AND DISCUSSION

Zeugorchis megacystis is parasitic in the lungs and upper digestive tract of *Thamnophis sirtalis parietalis, T. radix haydeni,* and *T. sauritus proximus.*

Neorenifer lateriporus is parasitic in the lungs and upper digestive tract of *Coluber constrictor flaviventris.*

Although differences of size were found between infected and noninfected snakes, both in males and in females, several factors complicate interpretation of these data: The relatively small number of infected snakes, the limited period in which they were collected, and the effects of bearing young. It is uncertain that differences in length-weight ratios can be attributed to parasitism.

Flukes were found in the mouth and pharynx of snakes caught in spring and summer (but not in autumn) and in the lung region in autumn. Probably some, if not all, adult lung-flukes migrate to the lungs from the mouth and pharyngeal regions at the advent of the hibernation season of the snakes.

The geographical distribution of infected snakes in Kansas showed no significant difference from that of noninfected snakes.

The pond snails *Helisoma trivolvis* and *Physa hawni* were examined for intermediate stages of the life cycle of the two new species of flukes. None was found.

The amphibians considered most likely to serve as intermediate hosts for these flukes were those with geographic distributions corresponding most nearly with those of the host snakes. For this reason, I examined newly transformed and adult *Rana pipiens* from Douglas County for metacercariae, but found none.

Thanks go to the following for help: Miss Mary E. Larson, Dr. A. Byron Leonard, Dr. Kenneth B. Armitage, Dr. Henry S. Fitch, Dr. T. H. Eaton, and Dr. Frank B. Cross, of the University of Kansas, and to Dr. Raymond M. Cable, of Purdue University.

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PLATE 1

A. *Zeugorchis megacystis* n. sp. (dorsal view).
B. *Neorenifer lateriporus* n. sp. (dorsal view).
(a, b, c, d—represent cross section levels of Plate 2).

ac. acetabulum
ce. cecum
cp. cirrus pouch
e. esophagus
ep. excretory pore (in a higher plane)
g. genital pore
m. metraterm
o. ovary
os. oral sucker
p. pharnyx
t. testis
u. uterus
ub. urinary bladder
v. vitellaria

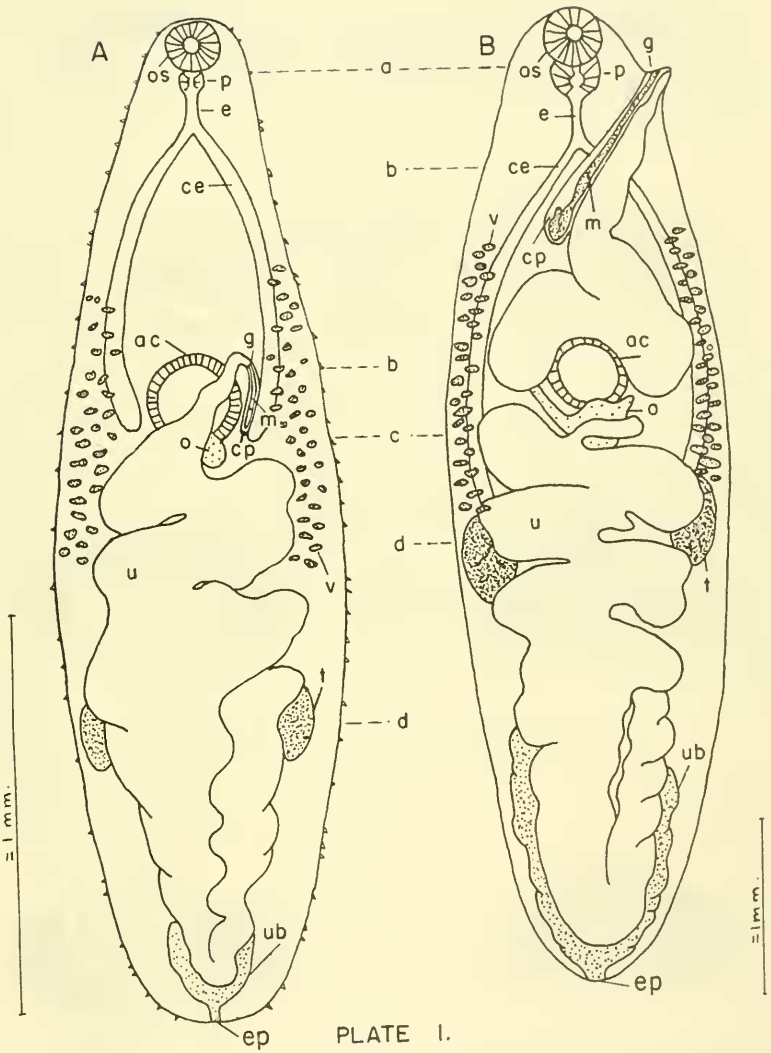

PLATE I.

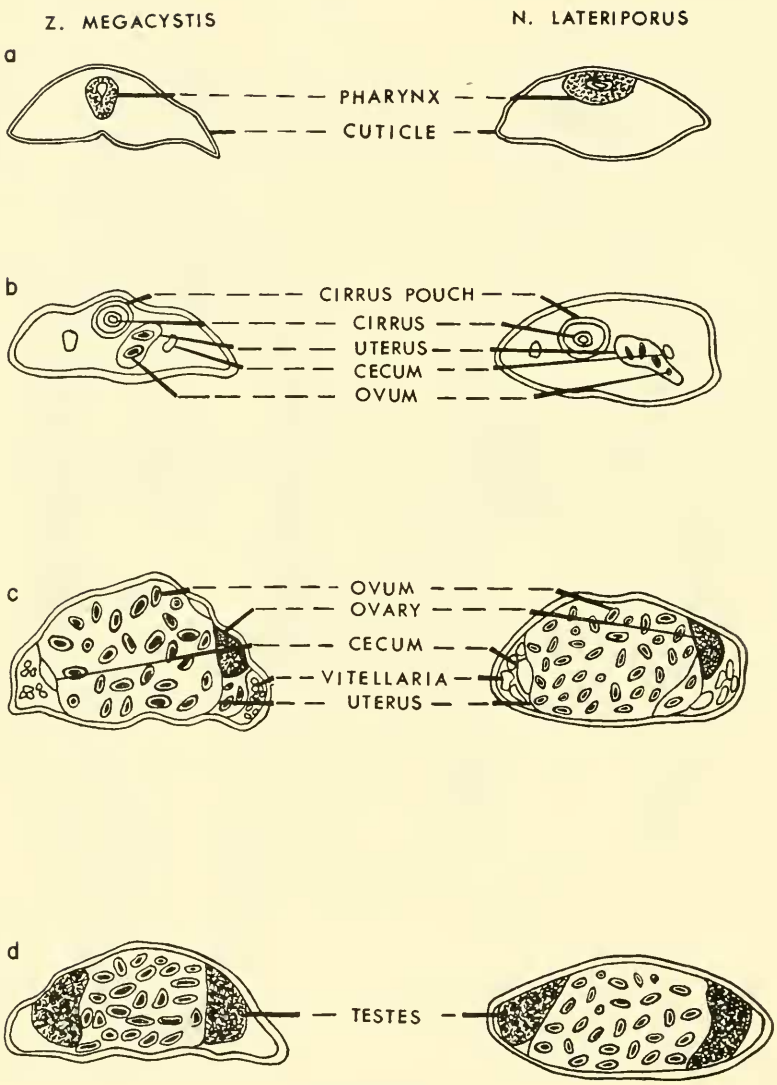

PLATE 2.

Cross section through *Zeugorchis megacystis* and *Neorenifer lateriporus*

a. Through pharnyx.
b. Through cirrus pouch.
c. Through ovary.
d. Through testes.